

Docket: NEB-20C2CD3

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

APPLICANTS: Wong-Madden, et al. EXAMINER:  
SERIAL NO.: Divisional Application of GROUP:  
U.S.S.N.: 08/560,809  
Filed: November 21, 1995  
FOR: Isolation And Composition Of Novel Glycosidases

The Honorable Commissioner of  
Patents And Trademarks  
Washington, DC 20231

Sir:

**PRELIMINARY AMENDMENT**  
**AND**  
**INFORMATION DISCLOSURE STATEMENT**

This is a Divisional Application of U.S. Application No. 08/560,809 filed November 21, 1995. Preliminary to examination on the merits, please amend the Application as follows:

**IN THE SPECIFICATION**

On page 1, line 4, please insert:

**--RELATED APPLICATIONS**

This is a Divisional Application of U.S. Application No. 08/560,809 filed on 21 November 1995 which is a CIP of 08/596,250 filed 24 June 1996 (Patent No. 5,770,405 issued on 23 June 1998) which is the National Stage of PCT US94/10758 filed on 22 September 1994 which is a CIP of 08/126,174 filed 23 September 1993 (now abandoned).--

Replace as-filed page 1 with substitute page 1.

**IN THE CLAIMS**

Please cancel claims 1-6 without prejudice.

Please add new claims 7-13

FOOTNOTES

7. (new) A method for modifying a carbohydrate, comprising the steps of:

- (a) selecting at least one glycosidase derived from *Xanthomonas*;
- (b) cleaving selected glycosidic bond between constituent monosaccharides of the carbohydrate by means of the glycosidase digestion; and
- (c) forming a modified carbohydrate.

8. (new) The method according to claim 7, wherein the glycosidase of step (a) is an exoglycosidase, the exoglycosidase being selected from the group consisting of a fucosidase and a  $\beta$ -galactosidase.

9. (new) The method according to claim 7, wherein the modified carbohydrate of step (c) has biological properties which differ from the glycosidase derived from *Xanthomonas*.

10. (new) The method according to claim 9, wherein step (c) further comprises altering the immunogenic properties of a glycoprotein.

11. (new) The method according to claim 9, wherein step (b) further comprises cleaving Fuc $\alpha$ 1-2R linkage.

12. (new) A method of selectively cleaving glycosidic linkage in a carbohydrate substrate comprising the steps of:

- (a) selecting a glycosidase from *Xanthomonas* having a substrate specificity for the glycosidic linkage, wherein said glycosidase is selected from the group consisting of a fucosidase and a  $\beta$ -galactosidase;
- (b) permitting the glycosidase to react with the carbohydrate substrate; and
- (c) cleaving the carbohydrate substrate.

13. (new) The method according to claim 12, wherein step (c) further comprises cleaving the carbohydrate substrate at a terminal glycosidic linkage selected from the group consisting of Fuc $\alpha$ 1-2R and Gal $\beta$ 1-3.

A copy of the new claims 7-13 is attached. 37 C.F.R. §1.121(c)(1)(ii).

FOOTNOTES

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE  
ISOLATION AND COMPOSITION OF NOVEL GLYCOSIDASES

RELATED APPLICATIONS

This is a Divisional Application of U.S. Application No. 08/560,809 filed on 21 November 1995 which is a CIP of 08/596,250 filed 24 June 1996 (Patent No. 5,770,405 issued on 23 June 1998) which is the National Stage of PCT US94/10758 filed on 22 September 1994 which is a CIP of 08/126,174 filed 23 September 1993 (now abandoned).

TECHNICAL FIELD

The present invention relates to novel glycosidases and their uses.

BACKGROUND OF THE INVENTION

The recognition that carbohydrates play a key role in biological processes of living organisms has made their study of great importance for medicine and basic science. The understanding of carbohydrates has lagged behind that of other types of biological molecules because of the immense complexity and variety of these molecules and the lack of availability of analytic and synthetic tools that enable scientists to differentiate one form from another.

**Forms of carbohydrates in nature.**

In nature, carbohydrates exist as polymers known as polysaccharides, that consist of a series of monosaccharides that are covalently attached by glycosidic bonds to form both branched and linear macromolecules. In addition, polysaccharides or, more commonly, oligosaccharides may be coupled to macromolecules such as proteins or lipids to form glycoproteins or glycolipids. Unlike naturally occurring polysaccharides, the oligosaccharides associated with protein or lipid consist of a relatively small subset of monosaccharide types.

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## NEW CLAIMS 7-13

7. (new) A method for modifying a carbohydrate, comprising the steps of:
  - (a) selecting at least one glycosidase derived from *Xanthomonas*;
  - (b) cleaving selected glycosidic bond between constituent monosaccharides of the carbohydrate by means of the glycosidase digestion; and
  - (c) forming a modified carbohydrate.
8. (new) The method according to claim 7, wherein the glycosidase of step (a) is an exoglycosidase, the exoglycosidase being selected from the group consisting of a fucosidase and a  $\beta$ -galactosidase.
9. (new) The method according to claim 7, wherein the modified carbohydrate of step (c) has biological properties which differ from the glycosidase derived from *Xanthomonas*.
10. (new) The method according to claim 9, wherein step (c) further comprises altering the immunogenic properties of a glycoprotein.
11. (new) The method according to claim 9, wherein step (b) further comprises cleaving Fuc $\alpha$ 1-2R linkage.
12. (new) A method of selectively cleaving glycosidic linkage in a carbohydrate substrate comprising the steps of:
  - (a) selecting a glycosidase from *Xanthomonas* having a substrate specificity for the glycosidic linkage, wherein said glycosidase is selected from the group consisting of a fucosidase and a  $\beta$ -galactosidase;
  - (b) permitting the glycosidase to react with the carbohydrate substrate; and
  - (c) cleaving the carbohydrate substrate.
13. (new) The method according to claim 12, wherein step (c) further comprises cleaving the carbohydrate substrate at a terminal glycosidic linkage selected from the group consisting of Fuc $\alpha$ 1-2R and Gal $\beta$ 1-3.